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Ataxin-2 polyQ expansions in FTLD-ALS spectrum disorders in Flanders-Belgian cohorts

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Abstract

There exists considerable clinical and pathological overlap between frontotemporal lobar degeneration (FTLD) and amyotrophic lateral sclerosis (ALS), which implies that these 2 neurodegenerative conditions share common pathogenic mechanisms. Recently, intermediate-length (27-33) polyglutamine (polyQ) expansions in ataxin-2 (*ATXN2*) have been associated with increased risk for ALS, while expansions of $>$ 34 repeats are known to cause spinocerebellar ataxia type 2 (Sca-2). We identified in 72 ALS patients one patient with a 33 polyQ expansion that was absent in 810 control individuals. This allele was also found in one patient with concomitant ALS-Sca-2. In contrast, in a Flanders-Belgian series of 270 FTLD and 22 FTLD-ALS patients, we found no association with intermediate-length polyQ expansions nor did we observe patient-specific long expansions in agreement with the recent observation in a screening of a substantial sized cohort of patients with diverse neurodegenerative brain diseases. Our results provide further support to the notion that *ATXN2* associated polyglutamine amplification is specific to the ALS-end of the FTLD-ALS disease spectrum.

Keywords: FTLD-ALS disease spectrum; Sca-2; Genetics; *ATXN2*; Genotype-phenotype correlations

1. Introduction

There exists considerable clinical, pathological and genetic overlap between frontotemporal lobar degeneration (FTLD) and amyotrophic lateral sclerosis (ALS). FTLD manifests clinically with progressive behavioral and language problems, which is complicated by the neuromuscular disorder ALS in 10–15% of patients (FTLD-ALS) (Lomen-Hoerth et al., 2010). In both FTLD and ALS, the majority of associated pathologies are characterized by neuronal and glial inclusions of the protein TAR DNA-binding protein 43 (TDP-43) (Neumann et al., 2006). Further, a common disease locus for FTLD and ALS has been identified at chromosome 9p21 (Morita et al., 2006;van Es et al.,

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Table 1
Characteristics of the patient and control cohorts

	FTLD (n = 270)	FTLD-ALS (n = 22)	ALS (n = 72)	Controls (n = 810)
Mean age O/I, SD, y	63.3 ± 10.4	59.6 ± 11.0	58.7 ± 12.3	65.1 ± 14.9
Male, n (%)	155 (57%)	9 (41%)	45 (63%)	345 (43%)
Familial, n (%)	90 (33%)	10 (46%)	18 (25%)	—
Known mutations, n (%)	15 (5.5%)[a]	0	2 (2.8%)[b]	—

[a] :13 *GRN* and 2 *VCP* mutations, [b] :1 *TARDBP* and 1 *FUS* mutation, O/I: onset/inclusion, SD: standard deviation

2009). Together, these findings have suggested that FTLD and ALS represent a continuum of neurodegenerative diseases and share common pathogenic mechanisms.

The default ataxin-2 (*ATXN2*) polyglutamine (polyQ) tract length is 22 to 23 repeats. Expansions of > 34 Q repeats cause autosomal dominant spinocerebellar ataxia type 2 (Sca-2) (Imbert et al., 1996) and in some patients familial Parkinson's disease (Gwinn-Hardy et al., 2000). Sca-2 is clinically characterized by deficits in motor coordination that affect gait, balance, speech and gaze. Recently, intermediate-length (27–33) polyglutamine expansions in *ATXN2* have been proposed as a novel risk factor for ALS (Elden et al., 2010). Most robust association has been found with longer polyQ lengths, 2 studies reported the 32 or 33 Q repeat expansions in ~1% of the ALS patients, whereas these alleles were not observed in control individuals (Elden et al., 2010;Lee et al., 2011). Further, it was shown that Sca-2 is also characterized by TDP-43 pathology, thereby providing a molecular link between Sca-2, ALS and FTLD (Elden et al., 2010). In this study we aimed to better assess the role of *ATXN2* polyQ expansions in the different phenotypes of the FTLD-ALS disease spectrum. Further, we describe the clinical features of 2 patients carrying a long intermediate-length polyQ expansion (33 Q repeats, Q^{33}).

2. Methods

Demographic descriptions of the patient and control study populations are summarized in Table 1. All participants were recruited with Ethical Committee approval and provided informed consent. Determination of the polyQ repeat length in *ATXN2* was performed as previously described (Elden et al., 2010). Two-tailed Fisher's exact test was used to evaluate genetic association. For a more detailed description of the study population and genetic analyses see Supplementary data.

3. Results

3.1. ATXN2 in FTLD-ALS spectrum disorders

In FTLD patients, the frequency of intermediate-length (27–33) polyQ expansions in *ATXN2* was similar as observed in control individuals (8/270 or 3.0% vs. 25/810 or 3.1%) (Table 2). The longest polyQ repeat length found in FTLD patients was 31 repeats, which was also observed in one control individual aged 76 years (Suppl. Table 1). In the 22 FTLD-ALS patients, one *ATXN2* intermediate-length polyQ repeat allele was observed (4.5%), with length of 27 Q repeats. On the other hand, our study population of ALS patients was found significantly enriched for *ATXN2* intermediate-length repeat carriers (7/72 or 9.7%, p = 0.012). The association was mainly driven by the longer (30–33) intermediate-length polyQ repeats, which were found in 4.2% (n = 3) of ALS patients, compared to 0.1% (n = 1) of healthy control individuals. One ALS patient was identified with a Q^{33} allele, which was absent in the 810 control individuals. During our analysis of the *ATXN2* gene, we identified a second patient with the Q^{33} allele who displayed ALS along with Sca-2. This patient was not included in the association analysis because of the mixed disease presentation.

3.2. Clinical phenotype of Q^{33} allele carriers

The first patient with the Q^{33} allele was a male patient with classical ALS. He presented with paresis of the left leg at the age of 69 years. His symptoms spread rapidly to include muscle weakness and pronounced fasciculations in all 4 limbs within a period of 6 months. At last medical examination he also showed mild hyperreflexia. Needle EMG confirmed neuropathic abnormalities in both legs and right arm. Normal sensory nerve action potentials were observed. The patient's history and neurological examina-

Table 2
The frequency of *ATXN2* polyQ expansions in FTLD, FTLD-ALS, ALS and control individuals

	≤ 26 Q repeats	27–33 Q repeats	> 31 Q repeats	*p*-value[a]
FTLD (n = 270)	262 (97.0%)	8 (3.0%)	0	ns
FTLD-ALS (n = 22)	21 (95.5%)	1 (4.5%)	0	ns
ALS (n = 72)	65 (90.3%)	7 (9.7%)	1 (1.4%)	0.012
Controls (n = 810)	785 (96.9%)	25 (3.1%)	0	

[a] Two-tailed Fisher's exact test

tion were unremarkable for cerebellar impairment. The movements in arms and legs were well coordinated, his speech was articulate and the eye movements were normal without ocular nystamus. The second patient with the Q^{33} allele had experienced slowly progressive gait disturbances starting from the age of 50 y. When evaluated at age 55, he showed a wide-based ataxic gait and mild muscular spasticity. Other examinations performed at that time were normal. At the age of 61 y, his clinical picture deteriorated rapidly with severe dysphagia and dysarthria. Atrophy and fasciculations in the tongue were seen. He showed strong overactive myotatic reflexes in the legs and arms and the plantar responses were extensor. Needle EMG testing revealed signs of active denervation in all extremities. The sensory nerve action potentials were of normal amplitude. MRI brain scan only showed mild cerebellar atrophy. The patient died of respiratory failure aged 63 y. Autopsy was not performed. His familial history was unremarkable for a movement disorder.

4. Discussion

The clinico-pathological overlap that exists between FTLD and ALS indicates that these 2 disorders share common pathogenic mechanisms. However, the precise molecular basis for this phenomenon remains enigmatic. In the present study we have investigated the role of the newly identified ALS gene *ATXN2* within the different phenotypes of the FTLD-ALS disease spectrum.

In our ALS sample, we confirmed association to *ATXN2* intermediate-length (27–33) polyQ expansions. Notably, we identified one ALS patient that carried a Q^{33} repeat allele, which was absent in the Flanders-Belgian or previously published control populations (Elden et al., 2010; Lee et al., 2011). The frequency of these high-risk alleles of $>$ 31 Q repeats in *ATXN2* is ~1% in our Flanders-Belgian ALS population, which is consistent with that of previously published results (Elden et al., 2010; Lee et al., 2011). This is comparable to the reported mutation frequency in the *FUS* and *TARDBP* genes in ALS patients (Mackenzie et al., 2010).

The commonly accepted cutoff for molecular diagnosis of Sca-2 is $>$ 34 Q repeats. Occasionally however, shorter alleles have been found in Sca-2 patients who developed the disease at a later age (Fernandez et al., 2000; Velázquez Pérez et al., 2009). In the present study, we identified the Q^{33} repeat allele in one patient with a pure, mainly lower motor neuron dominant, ALS phenotype. He displayed no signs of ataxia: the coordination of movements in his arms and legs, his speech and eye movements were all unremarkable. This was in contrast to the second Flanders-Belgian patient carrying Q^{33} who presented first with symptoms suggestive for a spinocerebellar disorder, but who several years later developed a rapidly progressive fatal motor neuron disease prompting a diagnosis of ALS. The rare association of Sca-2 and ALS has already been reported a few times before, but in patients who had longer *ATXN2* alleles (Infante et al., 2004; Nanetti et al., 2009). Taken together, these results indicate that significant overlap exists between the ALS and Sca-2 phenotypes in patients with intermediate-length polyQ expansions in *ATXN2* individuals may present with either ALS, Sca-2, or with overlapping phenotypes.

We found no genetic evidence that intermediate-length polyQ expansions in *ATXN2* also contribute to the etiology of FTLD or FTLD-ALS. The frequency of intermediate-length expansions (27–33) in *ATXN2* was not different between FTLD or FTLD-ALS patients and control individuals. Further, no patient-specific long expansions were observed in these patient cohorts. While our analysis of 270 FTLD patients was sufficiently powered, we cannot exclude that association or rare expansions of $>$ 31 Q repeats in *ATXN2* can be found in other large FTLD populations. However, our negative findings are in agreement with the recent publication of *ATXN2* association data that was obtained in a substantial sized cohort of patients with diverse neurodegenerative brain diseases including FTLD patients (Ross et al., 2011).

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Appendix. Supplementary data

Supplementary data associated with this article can be found, in the online version, at doi:10.1016/j.neurobiolaging.2011.09.025.

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